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Molecular Model of Extracellular Matrix Invasion by Cell Surface Activated Matrix Metalloproteinases

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Malignant cells exploit matrix metalloproteinases (MMPs) to promote extracellular matrix (ECM) invasion and metastasis. MMPs are zinc-dependent proteases that are secreted by mammalian cells as zymogens. Upon cell surface activation, they initiate tissue remodeling by catalyzing the proteolytic degradation of ECM macromolecules such as collagens and proteoglycans. The mechanism by which intact ECM is modified by MMPs remains unresolved. Specifically, it is unclear how these proteases remove physical barriers and process ECM components into substrates suitable for permeation. In addition, the ability of MMPs to degrade ECM components far away from the immediate pericellular environment of cells has been questioned due to the presence of natural MMP inhibitors in the vicinity of the cell surface that may block their potential long-range action. To provide new insights to these questions, we have developed a quantitative spectroscopic approach to intimately follow the pathways of ECM remodeling by Fourier transform infrared (FTIR) micro-spectroscopy. FTIR is an excellent technique for examining structural changes in proteins; due to the sensitivity of the Amide I band characteristics to the protein secondary structure. The combination of FTIR spectroscopy with a microscope provides a direct probe of the chemical composition of the intact matrix with a high spatial resolution. By utilizing a synchrotron IR source, it is possible to obtain chemical maps at a spatial resolution near the diffraction limit, which is 3-10 μm in the mid-infrared region. Thus, synchrotron-based FTIR micro-spectroscopy permits direct tracking of the degradation products along the enzymatic catalysis pathways on the biological matrix. Figure 1 shows the degradation pathways of matrigel matrix induced by cell secreted MMPs. The cells that were used in these studies are human fibrosarcoma HT-1080 that are characterized as highly invasive cancer cells. Our results show that the biological matrix proteolysis by "cell surface" activated MMPs is more pronounced in the invading front of the cell and it can proceed up to 30-40 μm around the pericellular environment of the cell. Interestingly, the degradation of the intact biological matrix occurs within distinct pathways or patches due to he complex morphology of the biological matrix. In addition, the net enzymatic proteolysis of the intact biologic matrix is rather "mild" and may be characterized mainly in destabilizing the local triple helices of the substrate molecules which promote the permeation of the ECM network, and enable the invasion of cells. The detected long range degradation of the ECM by MMPs suggest that the cell surface activation mechanism of MMPs and the inhibition process by their natural inhibitors may not be synchronized at the initial phase of MMPs expression by the cancer cells.

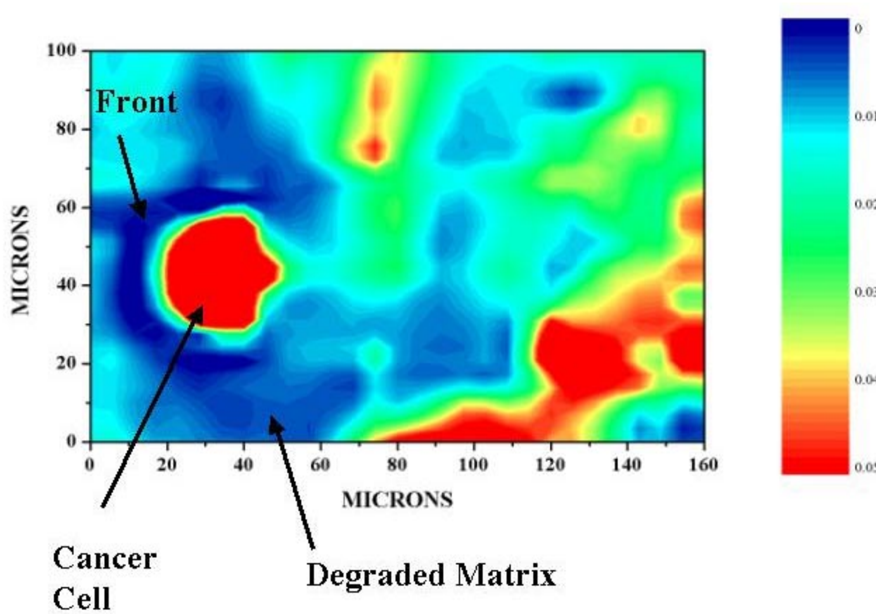

Figure 1. FTIR image of highly invasive human fibrosarcoma HT-1080 cell on matrigel matrix. The degradation pathways of the matrigel matrix by cell secreted MMPs were measure by FTIR micro-spectroscopy and the image was produced by correlating the degraded vs. the non-degraded spectra of matrigel where blue scores for highly degraded and red scores for non-degraded. The cell boundaries were verified by optical imaging and FTIR spectra of cell features.